buffer at 37° C. containing human PA28 activator (Boston Biochem, 12 nM final) with Ac-WLA-AMC (β5 selective substrate) (15 μM final), followed by 25 μL of assay buffer at 37° C. containing human 20S proteasome (Boston Biochem, 0.25 nM final). Assay buffer is composed of 20 mM HEPES, 0.5 mM EDTA and 0.01% BSA, pH7.4. The reaction is followed on a BMG Galaxy plate reader (37° C., excitation 380 nm, emission 460 nm, gain 20). Percent inhibition is calculated relative to 0% inhibition (DMSO) and 100% inhibition (10 μM bortezomib) controls.

[0101] When tested in this assay, compounds I-1 to I-21 all exhibited IC_{50} values less than 50 nM.

Example 3: Antiproliferation Assay

[0102] HCT-116 (1000) or other tumor cells in 100 μL of appropriate cell culture medium (McCoy's 5A for HCT-116, Invitrogen) supplemented with 10% fetal bovine serum (Invitrogen) are seeded in wells of a 96-well cell culture plate and incubated overnight at 37° C. Test compounds are added to the wells and the plates are incubated for 96 hours at 37° C. MTT or WST reagent (10 Roche) are added to each well and incubated for 4 hours at 37° C. as described by the manufacturer. For MTT the metabolized dye is solubilized overnight according to manufacturer's instructions (Roche). The optical density for each well is read at 595 nm (primary) and 690 nm (reference) for the MTT and 450 nm for the WST using a spectrophotometer (Molecular Devices). For the MTT the reference optical density values are subtracted from the values of the primary wavelength. Percent inhibition is calculated using the values from a DMSO control set to 100%.

Example 4: In Vivo Tumor Efficacy Model

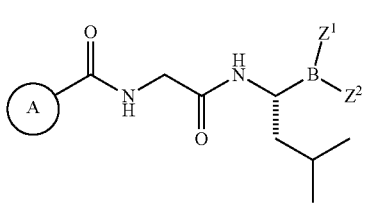
[0103] Freshly dissociated HCT-116 ($2\text{-}5\times10^6$) or other tumor cells in 100 μL of RPMI-1640 media (Sigma-Aldrich) are aseptically injected into the subcutaneous space in the right dorsal flank of female CD-1 nude mice (age 5-8 weeks, Charles River) using a 1 mL 26⅜-ga needle (Becton Dickinson Ref #309625). Alternatively, some xenograft models require the serial passaging of tumor fragments. In these cases, small fragments of tumor tissue (approximately 1 mm^3) are implanted subcutaneously in the right dorsal flank of anesthetized (3-5% isoflourane/oxygen mixture) C.B-17/SCID mice (age 5-8 weeks, Charles River) via a 13-ga trocar (Popper & Sons 7927). Beginning at day 7 after inoculation tumors are measured twice weekly using a vernier caliper. Tumor volumes are calculated using standard procedures ($0.5\times(\text{length}\times\text{width}^2)$). When the tumors reach a volume of approximately 200 mm^3 mice are randomized into treatment groups and begin receiving drug treatment. Dosing and schedules are determined for each experiment based on previous results obtained from pharmacokinetic/pharmacodynamic and maximum tolerated dose studies. The control group will receive vehicle without any drug. Typically, test compound (100-200 μL) is administered via intravenous (27-ga needle), oral (20-ga gavage needle) or subcutaneous (27-ga needle) routes at various doses and schedules. Tumor size and body weight are measured twice a week and the study is terminated when the control tumors reach approximately 2000 mm^3.

[0104] While the foregoing invention has been described in some detail for purposes of clarity and understanding, these particular embodiments are to be considered as illustrative and not restrictive. It will be appreciated by one skilled in the art from a reading of this disclosure that various changes in form and detail can be made without departing from the true scope of the invention, which is to be defined by the appended claims rather than by the specific embodiments.

[0105] The patent and scientific literature referred to herein establishes knowledge that is available to those with skill in the art. Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs. The issued patents, applications, and references that are cited herein are hereby incorporated by reference to the same extent as if each was specifically and individually indicated to be incorporated by reference. In the case of inconsistencies, the present disclosure, including definitions, will control.

What is claimed is:

1. A method for inhibiting proteasome activity comprising administrating to a patient in need of such inhibition a compound of formula (I), or a pharmaceutical composition comprising a compound of formula (I)



or a pharmaceutically acceptable salt or a boronic acid anhydride thereof, wherein Ring A is selected from the group consisting of